



United States Department of the Interior
U. S. GEOLOGICAL SURVEY
Columbia Environmental Research Center
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Date: April 1, 2013

To: Ed Hammer, USEPA Region 5, Chicago, IL

From: Chris Ingersoll and Ed Little

Subject: USGS Columbia Environmental Research Center (USGS-Columbia) quarterly project summary for the project entitled: "Protectiveness of water or sediment quality guidelines to species of special concern"

Attached please find the 01/01/13 to 03/31/13 quarterly project summary for USGS-Columbia project entitled: "Protectiveness of water or sediment quality guidelines to species of special concern." Please contact us if you have any questions concerning the attached summary (573/876-1819, fax -1896, email cingersoll@usgs.gov, elittle@usgs.gov). Please let us know if there are other individuals that should be receiving our quarterly summaries.

This quarterly summary provides progress on the following 5 tasks associated with the project:

- A. Task 1. Determining the acute and chronic toxicity of ammonia, nitrate and nitrite to amphibians (USGS lead investigator: Ed Little)
- B. Task 2. Developing and demonstrating a sediment toxicity test method with freshwater mussels for assessing sediment contaminants in the Great Lakes Basin and within North America (USGS lead investigator: Chris Ingersoll)
- C. Task 3. Evaluating of conditions used to culture or conduct toxicity tests with the amphipod *Hyalella azteca* (USGS lead investigator: Chris Ingersoll)
- D. Task 4. Assessing the toxicity of sulfate in water to early life stages of fish, snails, and mussels (USGS lead investigator: Ning Wang)
- E. Task 5. Improving concordance of sediment chemistry and toxicity (technical assistance to the USEPA Great Lakes National Program Office; USGS lead investigator: Chris Ingersoll)
- F. Task 6. Water-only toxicity testing of snails and mussels (USGS lead investigator: Ning Wang)
- G. Task 7. Major ion toxicity testing with select toxicity organisms (A: Influence of water hardness on the chronic toxicity of sulfate and chloride to fathead minnows (*Pimephales promelas*) and cladocerans (*Ceriodaphnia dubia*) and B: Acute and chronic toxicity of potassium to select aquatic organisms) USGS lead investigators: Ning Wang and Chris Ingersoll)
- H. Task 8. Water-only toxicity testing with sculpin and darters (USGS lead investigator: John Besser; Task not funded by EPA, but being conducted by USGS support in ongoing

GLRI studies)

cc: Ning Wang, Nile Kemble, John Besser, Bethany Williams, Carl Orazio, Rip Shively, Norman Grannemann (USGS)
Task 1: Chuck Stephan
Task 2 and 6: Chris Barnhart, Missouri State University
Task 3: Dave Mount, Russ Hockett, Chuck Stephan (USEPA), Lisa Taylor, Warren Norwood (Environment Canada), Dave Soucek (INHS)
Task 4, 7, 8: Chuck Stephan (USEPA), Dave Soucek (INHS), Mike Coffee (USFWS)
Task 5: Scott Ireland, Dave Mount (USEPA)

COLUMBIA ENVIRONMENTAL RESEARCH CENTER
UNITED STATES GEOLOGICAL SURVEY, COLUMBIA, MO
QUARTERLY PROGRESS SUMMARY

USGS Basis+ Program Element 42100, USGS Basis+ Project 2145E52 (USGS template/task 240 GLNPO sediment; USGS template/task 251 Hyalella; USGS template/task 252 Sulfate; USGS template/task 253 Mussel sediment; USGS template/task 254: Amphibians; USGS template/task 147 Major ion toxicity)

USGS Project Managers: Chris Ingersoll and Ed Little

Title: Protectiveness of water or sediment quality guidelines to species of special concern

USGS-Columbia is developing quarterly summaries for the USEPA that are intended to describe the status of activities associated with the seven following tasks associated with collaborative research between USGS and USEPA to evaluate the protectiveness of water or sediment quality guidelines to species of special concern (Interagency agreement between USEPA and USGS dated February 1, 2010).

- A. Task 1. Determining the acute and chronic toxicity of ammonia, nitrate and nitrite to amphibians (USGS lead investigator: Ed Little)
- B. Task 2. Developing and demonstrating a sediment toxicity test method with freshwater mussels for assessing sediment contaminants in the Great Lakes Basin and within North America (USGS lead investigator: Chris Ingersoll)
- C. Task 3. Evaluating of conditions used to culture or conduct toxicity tests with the amphipod *Hyalella azteca* (USGS lead investigator: Chris Ingersoll)
- D. Task 4. Assessing the toxicity of sulfate in water to early life stages of fish, and mussels (USGS lead investigator: Ning Wang)
- E. Task 5. Improving concordance of sediment chemistry and toxicity (technical assistance to the USEPA Great Lakes National Program Office; USGS lead investigator: Chris Ingersoll)
- F. Task 6. Water-only toxicity testing of snails and mussels (USGS lead investigator: Ning Wang)
- G. Task 7. Major ion toxicity testing with select aquatic organisms (A: Influence of water

hardness on the chronic toxicity of sulfate or chloride to fathead minnows (*Pimephales promelas*) and cladocerans (*Ceriodaphnia dubia*) and B: Acute and chronic toxicity of potassium or calcium to select aquatic organisms) USGS lead investigators: Ning Wang and Chris Ingersoll)

- H. Task 8. Water-only toxicity testing with sculpin and darters (USGS lead investigator: John Besser; Task not funded by EPA, but being conducted by USGS support in ongoing GLRI studies)

1. What work was accomplished for this past quarter (01/01/13 to 03/31/13)?

- A. Task 1. Determining the acute and chronic toxicity of ammonia, nitrate and nitrite to amphibians
1. Completed acute (96-hour) exposures to chloride (as NaCl) with both southern two-lined salamanders (*Eurycea cirrigera*) and mudpuppies (*Necturus maculosus*), indicating that later-stage larvae of these species can successfully be tested under static renewal conditions. Began data summary and analysis for these tests.
 2. Requested about 1000 mudpuppy larvae from Steve Redman of the Upper Midwest Environmental Sciences Center (La Crosse, WI) for testing in 2013. Collections to be initiated in April 2013.
 3. Scheduled conference call (1/15/13) to evaluate potential sites for amphibian tests associated with NAWQA Cycle III Regional Synoptic Study and to review study plan.
 4. Met with NAWQA team to coordinate 2013 studies.
- B. Task 2. Developing and demonstrating a sediment toxicity test method with freshwater mussels for assessing sediment contaminants in the Great Lakes Basin and within North America
1. USGS personnel participated in scheduled conference calls with USEPA and other interested groups to discuss the status of research conducted and planned associated with this task.
 2. Delayed the start of sediment toxicity study planned with fatmucket, midge, and amphipods exposed to dilution(s) of a highly contaminated sediment collected under the direction of Scott Ireland from the East Branch of the Grand Calumet River (Task 5). The reason for this delay was that preliminary studies were conducted with midge during the last quarter evaluating the control response (in water or in sediment) of 1-d-old vs. 4-d-old midge. A complimentary study is planned for the next quarter comparing control responses of amphipods (4- vs. 7-day old amphipods) measuring survival, weight, biomass, or reproduction.
 3. No additional exposures were conducted in the mussel sediment exposures because there were no species of the desired age or size available for testing during the past quarter.
- C. Task 3. Evaluating of conditions used to culture or conduct toxicity tests with the amphipod *Hyalomma azteca*
1. USGS personnel participated in scheduled conference calls with USEPA and other interested groups to discuss the status of research conducted and planned associated with this task and planned revisions to USEPA and ASTM methods based on the findings from these studies.
 2. A study was designed to evaluate the influence of age or feeding on amphipods in

chronic water or sediment exposures with control water or with control sediment was delayed until EPA Duluth completes preliminary amphipod feeding studies with diatoms.

- D. Task 4. Assessing the toxicity of sulfate in water to early life stages of fish, snails, and mussels
 - 1. See Task 7.
- E. Task 5. Improving concordance of sediment chemistry and toxicity (technical assistance to the USEPA Great Lakes National Program Office)
 - 1. USGS personnel participated in scheduled conference calls with USEPA and other interested groups to discuss the status of research conducted and planned associated with this task and planned revisions to USEPA and ASTM methods based on the findings from these studies.
 - 2. USGS developed a template for revising text, tables, and figures in the USEPA (2000) sediment toxicity methods manual. This template will be used to develop revised guidance for conducting 10-d and chronic amphipod and midge exposures described in the USEPA methods and for the ASTM methods.
 - 3. Conducted a study comparing the control response (in water or in sediment) of midge started with <1-h-old larvae compared to 4-d-old larvae. Larval survival, weight, and biomass was better with exposures started with 4-d-old larvae compared to exposures started with <1-h-old larvae. A companion study is planned with amphipods (starting with 4-d old vs. 7-d-old amphipods) evaluating survival, growth, and reproduction with different diets (e.g., YCT vs. diatoms).
 - 4. Results of these studies will be used to conduct a study to evaluate the response of different life stages of amphipods or midge (e.g., <24-h, 4-d vs 7-day old organisms) and mussels with exposure to dilutions of a highly contaminated and toxic sediment sample collected from the East Branch of the Grand Calumet River. The objective of this next study will be to determine if exposures started with 4-d-old midge improves control performance or if exposures started with 4-d-old amphipods will result in a delay of reproduction past Day 28 of a sediment exposure. Improved feeding methods in water exposures started with about 7-d-old amphipods has resulted in improved growth and reproduction starting before Day 28. Hence, for sediment exposures with amphipods where reproduction is to be determined, it may be desirable to start with younger amphipods (4-d-old rather than 7-d-old) to delay the onset of reproduction past the end of the 28-d sediment exposure.
- F. Task 6. Water-only toxicity testing of snails and mussels (USGS lead investigator: Ning Wang)
 - 1. Searched Tier 1 chemical toxicity data for commonly tested amphipods, cladocerans, and snails in literature, and established a range of exposure concentrations of the Tier 1 chemicals to two commonly test organisms (*Hyalella* and *Ceriodaphnia*) and two snails.
- G. Task 7. Major ion toxicity to select aquatic organisms (USGS lead investigators: Ning Wang and Chris Ingersoll).
 - 1. Task 7a: Sulfate or chloride toxicity influenced by water quality
 - a. Completed data analysis for a short-term 14-day static-renewal toxicity test with fathead minnows starting with newly fertilized eggs in three test water

(CERC 100 mg/L hard water, the 100 mg/L hard water with addition of chloride to 25 mg/L, and ASTM moderately hard reconstituted water).

- b. A reconstituted water was designed to match the water quality characteristics of ASTM moderately hard water and prepared by diluting the CERC well water with deionized water, with additional K_2SO_4 , Na_2SO_4 , and $MgSO_4$. This reconstituted water is planned to be used for determine why fathead minnow are sensitive to sulfate in diluted well water but not in ASTM water. The results of chemical analyses for the new water indicate the hardness, pH and major ions were close to those in the ASTM moderately hard water.
- c. Completed a 90-day NaCl exposure of fatmucket. Treatments included 1.0, 0.5, 0.25, 0.13, and 0.063 g NaCl/L with or without the presence of a thin sand substrate. Mean control survival was >98% on Day 28, >83% on Day 56 in both the water-only and sand treatments. The control survival reduced to 53% at the end of 90-day sand treatment (the water-only tests ended on Day 60). While survival may not be significantly reduced with exposure to the highest concentration of NaCl, growth (shell length) was visually observed to be reduced in the highest concentration of NaCl tested.
- d. Completed a memo to EPA Region 5 summarizing the results of 96-h NaCl toxicity tests conducted at our facility over the past several years.
- e. Completed a 24-h fatmucket glochidia test in five waters. The control viability was >95% in all tests. The EC50s between the two test waters (ASTM moderately hard water and 100 mg/L hard well water) are similar (around 700 mg Cl/L). Water hardness (50 to 200 mg/L) seems to influence the Cl toxicity: The EC50 is around 380 mg Cl/L at hardness 50 mg/L, around 700 mg Cl/L at hardness 100 mg/L, and around 1400 mg Cl/L at hardness 200 mg/L. However, the EC50 at hardness 300 mg/L is similar to the EC50 at hardness 200 mg/L.

2. Task 7b: Potassium or calcium toxicity influenced by water quality

- a. No testing was conducted in this past quarter.

H. Task 8. Water-only toxicity testing with sculpin and darters (USGS lead investigator: John Besser)

- 1. No additional toxicity testing was conducted with sculpin or darters during the past quarter.

2. What problems (or sources of error) were encountered, if any?

None

3. If a problem was encountered, what action was taken to correct it?

Not applicable

4. What work is projected for the new quarterly activity?

- A. Task 1. Determining the acute and chronic toxicity of ammonia, nitrate and nitrite to amphibians

1. Continue summarizing data from wood frog and gray tree frog embryos exposed to NaCl, KCl, CaCl₂, and Na₂SO₄ in 2012 tests.
 2. Participate in conference calls with USEPA to plan potential test designs for 2013 definitive tests with southern two-lined salamander larvae (*Eurycea cirrigera*) and/or mudpuppies (*Necturus maculosus*). Discuss the possibilities for testing earlier-stage larvae to evaluate life-stage-specific differences in sensitivity seen in previous tests with gray treefrogs (*Hyla versicolor*) and wood frogs (*Lithobates sylvaticus*).
 3. Procure muddy puppies and initiate toxicity tests with them.
 4. Continue discussions about study design for atrazine/nutrient exposures of southern leopard frogs (*Lithobates sphenoccephalus*) or gray treefrogs (*Hyla versicolor*) in association with NAWQA Cycle III Regional Synoptic Study.
- B. Task 2. Developing and demonstrating a sediment toxicity test method with freshwater mussels for assessing sediment contaminants in the Great Lakes Basin and within North America
1. Continue evaluating behavior of various species of early life stages of mussels in various sediments using the exposure system described in the October 2012 quarterly summary.
 2. Identify additional sediments for evaluating mussel toxicity testing methods (e.g., sediment dilution study with East Branch Grand Calumet River sediment).
 3. Continue to develop plans for coordinating sediment testing of mussels as part of the USGS NAWQA agriculture project planned for the Midwest in 2013. This USGS NAWQA project will include sediment toxicity tests conducted with *H. azteca* or *C. dilutus* (information was provided to USEPA in an email dated September 28, 2012). The plan would be to test mussels in about 50 of the 100 sediments to be evaluated in 2013 by the USGS NAWQA program.
- C. Task 3. Evaluating of conditions used to culture or conduct toxicity tests with the amphipod *Hyaella azteca*
1. Continue summarizing data for the four 42-d water-only test to evaluate the influence bromide or iodide on the response of *H. azteca* in reconstituted waters.
 2. Continue developing plan with the HAAG for conducting inter-laboratory testing of the water-only *H. azteca* toxicity testing method.
 3. Continue evaluating methods for culturing and testing known-age amphipod and midge (e.g., <24-h old, 4-d old, 7-d old).
 4. Conduct study to evaluate starting age of *H. azteca* in control water or in control sediment using different diets.
- D. Task 4. Assessing the toxicity of sulfate in water to early life stages of fish, snails, and mussels
1. See Task 7.
- E. Task 5. Improving concordance of sediment chemistry and toxicity
1. Continue discussions with USEPA GLNPO regarding ongoing and future options for technical assistance associated with this task (e.g., additional sediments, comparisons of diets for *H. azteca* or *C. dilutus*, evaluate starting age of *H. azteca* in water or in sediment toxicity test [e.g., starting exposures with <24-h old or 4-d-old organisms, rather than 7-day old organisms]).
 2. Continue revisions to USEPA and ASTM standard methods for conducting 10-d and chronic sediment and water toxicity tests with *H. azteca* or *C. dilutus*.

3. Plan for study to conduct additional testing of diluted toxic sediment to evaluate starting age or diet on responses of amphipods or midge in chronic sediment exposures.
 - F. Task 6. Water-only toxicity testing of snails and mussels
 1. Begin Tier 1 toxicity testing with two commonly test organisms (*Ceriodaphnia dubia*, *Hyalella azteca*) and with snails (*Lymnaea stagnalis*, *Fluminicola* sp., or *Physa gyrina*) cultured in the CERC.
 2. Continue to work with chemists and others who have used the Tier 2 chemicals in their toxicity tests, to find the methods to maintain more constant exposure concentrations for some of these chemicals, such as, preparing stock of organic chemicals in solvent, “equilibrating” test chambers with the test material before starting the test to minimize the sorption of organic chemicals to the surface of test chambers, and conducting aluminum toxicity test with water at pH 6.
 3. Continue compiling toxicity databases and associated toxicity thresholds for Tier 1 and Tier 2 chemicals (e.g., final acute values).
 - G. Task 7. Major ion toxicity to select aquatic organisms (Task 7a: and Task 7b:)
 1. Task 7a: Sulfate or chloride toxicity influenced by water quality
 - a. Continuously discuss with the USEPA Region 5 and Dave Mount (USEPA) and Dave Soucek (Illinois Natural History Survey) to design short-term sulfate toxicity test with fathead minnows, and begin another 14-day fathead minnow sulfate toxicity test in three test waters, including 100 hard diluted well water, ASTM moderately hard water, and diluted well water adjusted to water quality characteristics of the ASTM water.
 - b. Measure shell length and dry weight of surviving mussels in the 90-day NaCl tests with fatmucket.
 - c. Analyze the 24-hour fatmucket glochidia NaCl test data.
 - d. Continue discussions with Dave Mount and Dave Soucek on what water quality variables we might want to evaluate in additional NaCl exposures.
 - e. Continue compiling toxicity databases and associated toxicity thresholds (e.g., final acute values) for sulfate and chloride.
 2. Task 7b: Potassium or calcium toxicity influenced by water quality
 - a. Conduct 48- or 96-h KCl and CaCl₂ toxicity data with *Ceriodaphnia dubia*, *Hyalella azteca* and with two snails (*Lymnaea stagnalis*, *Fluminicola* sp., or *Physa gyrina*) cultured in the CERC.
 - b. Continue discussions with Dave Mount and Dave Soucek on what water quality variables we might want to evaluate in additional KCl or CaCl₂ exposures.
 - c. Continue compiling toxicity databases and associated toxicity thresholds (e.g., final acute values) for KCl and for calcium.
 - H. Task 8. Water-only toxicity testing with sculpin and darters
 1. No toxicity testing of sculpin or darters are planned for the next quarter.
5. Is the project work on schedule?

For the quarter? Yes

For the project? Yes

6. Does the project funding rate support the work progress?

Yes, given the revised budget period for Task 7 will continue to through June 2016.

7. What has been spent to date?

If requested, USGS will provide USEPA (or other interested groups) summaries of expenditures on the project.

8. Have you submitted a quarterly voucher for reimbursement?

USGS will provide vouchers as necessary.

9. Is there a change in principal investigator?

No

10. If you have a multi-year project with budget periods, have you submitted your request for a funding amendment?

Not applicable